DISTRIBUTION OF IRON IN THE TISSUES OF SOME BIVALVE MOLLUSCS

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ABSTRACT

The quantitative distribution of iron in the tissues has been studied in three species of bivalves. Two of these, Arca granosa and Arca inaequivalvis possess haemoglobin in their blood corpuscles, whereas the third species, Meretrix casta, which has been investigated for comparison, does not possess haemoglobin. The concentration of iron in the digestive diverticula of Arca is considerably higher than in the other tissues and also than that in the digestive gland of Meretrix. The relatively high concentration of iron in the digestive diverticula is similar to that in the haemopoietic organs of vertebrates. This would suggest that the digestive diverticula of Arca have possibly a role similar to that of the haemopoietic organs.

INTRODUCTION

Iron is micro-constituent of all living tissues. Its micro-concentration generally varies from 0.01 to 0.10 mg. per gm. of cell substances (Fearon, 1948). It is also an invariable secondary element in all red-blooded animals. In the marine biosphere the concentration of heavy metals is significantly higher than in the hydrosphere (Cornec, 1919). With notable exceptions, such as copper in haemocyanin and iron in haemoglobin, the trace elements found in many invertebrates have no known function. Vinogradov (1953) has compiled data on the elementary chemical composition of living organisms, but much of the work referred to by him was conducted long ago by experimental techniques which would not be regarded now as valid. Recently, Galtsoff (1964) investigated in Crassostrea virginica (Gmelin) the seasonal variation in iron content. Brooks (1965) investigated the biogeochemistry of trace element uptake by some New Zealand bivalves. Hobden (1967) has studied the iron metabolism in Mytilus edulis. Partly in view of the relative paucity of information on the distribution of iron in marine
animals, the present study was made on the distribution of iron in the
tissues of bivalve molluscs, *Arca granosa* (Linne), *Arca inaequivalvis* (Bru-
guier). A more important justification is that *Arca* is one of the very few
bivalves, which have haemoglobin contained in blood corpuscles. The
tissues of *Meretrix casta* (Chemnitz), which have no haemoglobin, have
also been studied for comparison.

**Material and Methods**

Freshly collected bivalves were cleaned and kept in filtered sea-water
for two days before being used, so that the undigested food materials might
be eliminated from the intestine.

A stainless steel scalpel was used to cut the adductors of the animals
in a sample. The following tissues were isolated: mantle, gills, foot, ad-
ductor muscle and digestive diverticula. They were washed well in distilled
water and dried between the folds of filter-paper and weighed. For each
estimation tissues from eight animals were pooled together. The tissues
were digested in H$_2$SO$_4$. An equal quantity of saturated potassium per-
sulphate was added and the mixture left overnight. The solution was then
diluted with distilled water and a few drops of 10% sodium tungstate added
to precipitate protein. The solution was then filtered and made up to a
known volume. Five ml. of 2M potassium thiocyanate was added to
5 ml. of this solution and the optical density at 480 mµ measured imme-
diately. Standard solution was prepared from freshly prepared solution of
ferric ammonium sulphate. Following the suggestion of Collins and Diehl
(1959) the glassware other than digestion flasks was not subjected to acid
cleaning.

The data relating to iron content in different tissues were statistically
tested. The values were found to be significant at 5% level.

**Histochemistry**

Standard histochemical techniques were used to locate iron within the
cells and tissues. Tissues were fixed in 10% neutral formalin embedded
in wax and cut into sections 10 mµ thick. Dewaxed sections were treated
with 2% sodium ferrocyanide for 5 minutes, then with a mixture of equal
volumes of 2% sodium ferrocyanide and 2% hydrochloric acid for 1 hour.
This solution was renewed after 30 minutes. Some sections were counter-
stained with eosin. Final mounting was with Canada balsam.